Modern trends in Biological Sciences

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Pectin: Extraction from stem pith of banana and structural and rheological characterisation

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Abstract

Pectin is extracted from the stem pith of Poovan variety of Banana (*Musa sp*.). It was found to contain a good amount of extractable pectin. The tested range of pH and Solid-Liquid Ratio were found to be affecting the yield of pectin slightly, while an increase in temperature and duration of heating were found to be increasing the yield. The pectin obtained was found to High Methyl pectin. Yield as calcium pectate was 57.87%. FTIR and NMR spectra of the pectin obtained indicate that it contains Galacturonic acid, galactose, rhamnose, and arabinose. Both β1 - 4 and β 1 - 6 glycosidic linkages were found between galactose residues while galacturonic acid was found to be linked to rhamnose by α1 - 2 glycosidic bond. All the above details are discussed in this article.

Keywords: Pectin, Value added products, Agricultural Wastes

Abbreviations: DM: the degree of methylation, HM pectin: high methyl pectin, SLR: Solid to the Liquid ratio

Introduction

Recently, agriculture in India is running into a loss for farmers, primarily because of the increased cost of production and decreased yield due to biotic and abiotic factors, coupled with competition in international markets and the resulting decrease in the price of the product. Therefore, nowadays, educated youth hesitates to take up agriculture as a major source of income. At the same time, agricultural land is used for some other purposes, which has affected the climatic conditions of our country. This problem can be solved to a great extent by producing alternate value-added products from crop plants. India, being the largest producer of banana (*Musa sp*.) in the world, has tremendous potential for producing such non-conventional products such as pectin from this kind of crops.

According to Horticulture Statistical Division 2017[1], banana is the major fruit crop produced in India (33.4% of the total fruit production in 2013-14) with a total production of 29.163 million tonnes from over 8 lakh hectares of land. India is the largest producer of banana in the world, producing 24.5% of the total global production in 2014. Only the fruits of the plant are used while the bulk of the remaining plant body is treated as waste. If these waste materials are used for the production of pectin by simple cost-effective means, it will make cultivation of banana more profitable by increasing the income of the farming community. At the same time, it will create more employment opportunities in the agriculture-related industries, and also may solve the problem of pollution due to the decomposition of waste materials. Moreover, every year a major share of the banana plants is lost/damaged even before harvesting due to rain, wind, pest infestation and other climatic/environmental adversities. If these damaged plant parts are used for the production of pectin, the confidence of farmers can be boosted to a great extend even at these extreme situations.

Pectin is a natural hetero-polysaccharide, consisting mainly of galacturonic acid residues, along with varying amounts of rhamnose, arabinose, galactose etc.[2]. It commonly occurs in the cell wall and middle lamella of all land plants. The carboxylic group of galacturonic acid residue may be randomly esterified with methanol while C-2 and/or C-3 may be O-acetylated. The degree of methylation, expressed as the percentage of methylated galacturonic acid residues, is a very important parameter in deciding the chemical structure and properties and therefore the application of the pectin extracted. The ultimate structure of pectin depends upon the source and conditions of extraction[3].

Pectin is used as a gelling agent in confectionary/cosmetic/food industries and is also found to have clinical and pharmaceutical uses. Being a soluble dietary fiber that is fermented in the human colon by various flora, it may also be used as a food supplement[2]. Gel formation as well as the strength of the gel formed, are promoted by molecular weight and also by the concentration of pectin used. The fine structure of pectins may be modified to suit the variety of uses in the industries. At the same time, genetic modification of particular plants may be used to produce pectin with special properties[4].

It is well known that pectin has a cholesterol-lowering effect and is dependent on its ability to form a viscous gel[5]. Pectin is thought to be achieving this by binding to cholesterol and bile acids, thereby preventing their reabsorption from the gut[6],[7]. Pectin may also be involved in preventing the formation of the micelle and the absorption of cholesterol[8].

It is reported that the effects of dietary pectin against colon cancer are directly or indirectly related to its probiotic activity. Both pectic oligosaccharides and modified pectin derivatives are reported to inhibit the growth of harmful colon microbes[9,10]. Heat-modified Citrus pectin has demonstrated to induce apoptosis of androgen-responsive and androgen-independent prostate cancer cells[11]. Some studies indicate that pectin from apple or citrus can decrease the activity of β-glucuronidase, β-glucosidase and tryptophanase formed in the alimentary canal from the microbes[12]. It is believed that these enzymes have a role in the initiation of carcinogenesis.

Some workers have reported that, when nicin (antibacterial natural food bio preservative) is encapsulated with pectin-nicin composite, its antimicrobial activity increased[13]. They also found that as the Degree of Esterification (DE) of the pectin increased, the antimicrobial activity decreased.

It was reported earlier that modified citrus pectin can inhibit tumour growth, angiogenesis and metastasis[14]. Amidated low methyl pectin can be prepared from pectin by treatment with ammonia[15]. In another report, different cations were used to study the gel microstructure and found that the type of cation along with degree of methylation and source of pectin influenced the gel microstructure[16]. Similarly, the incorporation of phenolic compounds in to the pectin moiety was found to be increasing the hydrophobicity and antioxidant property of the modified pectin[17]. Pectin may be oxidised using ferulic acid units to produce cross-linked pectin, which has better rheological properties[18]. Thus, pectin can be modified in a variety of ways to suit our specific applications.

Since our understanding on the applications of pectin in various fields is increasing day-by-day, it is essential to explore the potential for the production of pectin from new, feasible sources. Also new insights in to the diversity of pectin organisation in various species may contribute to its increased applications.

Currently, pectin is commercially produced from apple pomace and citrus peel, both of which are not abundantly available in India.

This study is aimed at exploring the possibility of extraction of pectin from the stem pith of banana (*Musa sp.*), the effect of various conditions of extraction upon the yield of pectin and also the characterisation of the pectin obtained to certain extend. The various parameters being investigated are- SLR (Solid-Liquid Ratio – number of grams of the powdered stem pith to the number of milliliters of the extractant. The ranges studied are 1:30, 1:40, 1:50 and 1:60), pH of the extractant : (water having pH lowered with HCl; the ranges studied are 1.0, 1.5, 2.0, 2.5 and 3.0), Duration of heating: 30 minutes, 52.5 min, 75 min, 97.5 min and 120 min., Temperature at which extraction is carried out: 40, 54, 68, 82 and 96 ^{0}C.

Materials and methods

1. Material

All reagents are purchased from Merck, India unless otherwise specified.

1.1 Preparation of plant material

Stem pith of Poovan variety of banana (*Musa sp*.) was collected from a local farmer in Palakkad district, Kerala, India. It is then washed with a mild acid, chopped into small pieces, pulverized using a food processor, sun-dried to constant weight, powdered and kept in an airtight vessel for further use.

1.2. Preparation of extractant solution

Distilled water is mixed with hydrochloric acid to attain a particular pH as is shown in table- 1.

2. Extraction of crude pectin

10g of the powdered material is weighed (except for 5HA5, which was 8 g), tied up in a bag made of cotton cloth, immersed in a particular volume of water maintained at a pH and heated at a constant temperature (table 1) in a 500 – ml Erlenmeyer flask (Borosil) for a certain duration of time. After heating, the flask is cooled to room temperature, the bag containing powdered material was taken out and squeezed to release any drop of liquid. Then pH is brought above 6.0 using Barium Carbonate, centrifuged at 5000 rpm for 10 minutes and the supernatant is collected. It is then evaporated under vacuum to a volume of 50 ml, the double volume of isopropyl alcohol is added, shaken well and kept at room temperature for 24 hours. It is then centrifuged at 5000 rpm for 10 minutes, and the supernatant is discarded. The precipitated crude pectin is dried under air to constant weight, powdered and transferred to air-tight vials for further analysis[19].

Characterisation

3.1. Estimation as calcium pectate

The method described by Ranganna S. (1979) was used with some modifications[19].

0.735 g of the powdered extracted pectin was dispersed in 10 ml of 0.01 N HCl, kept in boiling water bath for 20 minutes, cooled to room temperature, centrifuged at 5000 rpm for 10 minutes, and the supernatant collected. The residue was mixed with 10 ml of 0.05 N HCl, kept in boiling water bath for 20 minutes, cooled to room temperature, centrifuged at 5000 rpm for 10 minutes, and the supernatant collected. The residue was mixed with 10 ml of 0.3 N HCl, kept in boiling water bath for 20 minutes, cooled to room temperature, centrifuged at 5000 rpm for 10 minutes, and the supernatant collected. The residue was mixed with 10 ml of water, kept in boiling water bath for 20 minutes, cooled to room temperature, centrifuged at 5000 rpm for 10 minutes, and the supernatant collected.

All supernatants were pooled together and made up to 100 ml with water. 30 ml of this solution was pipetted out in to a 500 ml Erlenmeyer flask, 3 drops of phenolphthalein were added as indicator, neutralised with 1 N NaOH. An excess of 3.0 ml of 1N NaOH were added for saponification, shaken well and kept at room temperature for 48 hours. Then 3.0 ml of 1 N acetic acid was added to it, shaken well. After 15 minutes, 4.0 ml of 1N $CaCl_2$ was added with constant shaking and was allowed to stand for 4 hours. Then it was boiled for 2 minutes, filtered using a pre-weighed filter paper (HiMedia Laboratories) under vacuum, washed with hot water, tested with silver nitrate for the presence of chloride, dried in a vacuum desiccator for constant weight.

$$\% \text{ of calcium pectate} = \frac{W \times V1 \times 100}{V2 \times P}$$

where, W = weight of calcium pectate, V1 = total volume of solution prepared, V2 = volume of solution used for precipitation ,P = amount of pectin used

3.2. Estimation of Degree of Esterification (D.E.)

The method described in Food Chemicals Codex[20] is used with some modifications.

0.2 g of the powdered pectin is mixed with 5 ml of ethanol, then 45 ml of H_2O (HPLC) were added, mixed well, kept in a boiling water bath for 10 minutes, cooled to room temperature, centrifuged at 4500 rpm for 10 minutes, supernatant has been collected and made up to 100 ml with HPLC grade water. 10 ml of this extract is pipetted out into a conical flask, 2 drops of phenolphthalein added as an indicator, titrated against 0.01 N NaOH. Value noted as V1 and is the measure of the unesterified galacturonic acids. 5.0 ml of 0.1 N NaOH is added to the above solution, shaken well, and kept at room temperature for 3 hours for saponification. Then 5.0 ml of 0.1 N HCl is added to it to neutralize the NaOH, 2 drops of phenolphthalein added as an indicator, and titrated against 0.01 N NaOH, value noted as V2. This is the measure of the esterified galacturonic acid.

$$DE = V2 \times 100/V1 + V2$$

3.3. IR analysis:

The powdered pectin was mixed with KBr (1:100) and pressed into pellets. Then it is analysed with a Jasco-4100 (Japan) machine and FTIR spectra were collected at the transmission mode in the frequency range of 400-4000 cm^{-1}, resolution = 2 cm^{-1}, No. of scans = 8.

3.4. NMR analysis:

NMR spectra at 400MHz, used liquid state NMR spectrometer (Bruker AV 400) with D_2O as a solvent for the analysis.

Results and Discussion

Effect of various extraction conditions upon pectin yield

Table - 1 shows the yield under specified conditions of extraction (Values are averages of six independent analysis ± SEM)

a. Provided all other conditions of extraction being identical, yield has been increased as the duration of heating increased. This is evident from the following pairs of sets: 5HA1 and 5HA16, 5HA6 and 5HA7, 5HA28 and 5HA20, and 5HA24 and 5HA10. In all the above-mentioned cases, the percentage of increase in yield of pectin upon the increased duration of heating is noticeable.

b. As the temperature of extraction increases, yield also increases. This is evident from the relevant values of the following pairs of sets: 5HA1 and 5HA6, 5HA16 and 5HA7, 5HA11 and 5HA2, 5HA29 and 5HA30, 5HA15 and 5HA18, 5HA13 and 5HA21, and HA9 and 5HA10. At a pH of 1.5 and for a less time period of heating, the temperature was a very important limiting factor. This is evident from the sets 5HA1 and 5HA6, 5HA15 and

5HA18 all of which heated for 52.5 minutes. The first pair demonstrated an increase of 926 % while the second one, an increase of 1044.8 % because of the increase of temperature from 54^0C to 82^0C. However, when the mixtures were heated for a longer period of duration (97.5 min), the increase in the percentage of yield decreased to 163 % (5HA13 and 5HA21) or 728 % (5HA16 and 5HA7).

All these observations may be because as the duration and temperature of heating increases, more of the extractant penetrates the cellular matrix of the stem pith of banana, thereby releasing more pectin from chemical bonds with other cell wall components.

Set	SLR	pH	Time(min)	Temp(^{0}C)	Yield(%)
5HA1	30	1.5	52.5	54	1.77±0.34
5HA2	40	2	75	68	3.7±0.91
5HA3	30	2.5	52.5	82	3.73±0.27
5HA4	40	2	75	68	2.82±0.13
5HA5	60	2	75	68	4.33±0.4
5HA6	30	1.5	52.5	82	16.4±1.64
5HA7	30	1.5	97.5	82	27.5±0.7
5HA8	20	2	75	68	1.62±0.22
5HA9	50	2.5	97.5	54	1.23±0.16
5HA10	50	2.5	97.5	82	4.97±0.86
5HA11	40	2	75	40	0.97±0.18
5HA12	40	2	75	68	3.43±0.84
5HA13	50	1.5	97.5	54	15.1±0.67
5HA14	50	2.5	52.5	54	1.7±0.09
5HA15	50	1.5	52.5	54	2.68±0.47

5HA16	30	1.5	97.5	54	3.78±0.52
5HA17	40	2	75	68	7.7±1.37
5HA18	50	1.5	52.5	82	28±2.25
5HA19	40	1	75	68	5.47±0.78
5HA20	30	2.5	97.5	54	2.07±0.11
5HA21	50	1.5	97.5	82	24.6±2.46
5HA22	40	3	75	68	3.12±0.28
5HA23	40	2	120	68	8.05±1.53
5HA24	50	2.5	52.5	82	3.57±0.64
5HA25	30	2.5	97.5	82	2.93±0.06
5HA26	40	2	30	68	2.97±0.47
5HA27	40	2	75	68	5.23±0.79
5HA28	30	2.5	52.5	54	2.5±0.39
5HA29	40	2	75	68	7.48±1.16
5HA30	40	2	75	96	17.5±1.87

Table 1. The yield of pectin (% of dry weight) from stem pith of Banana

c. Yield drastically increases when pH decreases from 2.5 to 1.5. This is clear from the analysis of the following pairs of sets: 5HA6 and 5HA3, 5HA16 and 5HA20, 5HA7 and 5HA25, 5HA15 and 5HA14, 5HA18 and 5HA24, 5HA13 and 5HA9, 5HA21 and 5HA10. This may be because at the higher pH, the extractant may have lower penetrability into the cell wall materials and also because the chemical bonds between pectin and other cell wall components become weaker at the lower pH. At pH = 1, the yield was very little (5HA19), probably because the other factors (duration of heating and temperature) might not be in their optimal levels. Also, the higher concentration of the acid might have disintegrated the pectin released.

Increase in yield with a decrease in pH was noticeable in the sets in table 2. As is evident from the table, a feature common to all sets with increased yield is that they are maintained at a temperature of 82^{0}C. Even though 5HA13 demonstrated an incredible increase of 1227.6% in yield over 5HA9, the absolute yield of 5HA13 was lower (only 15.1%) probably because of its lower temperature of 54^{0}C.

Set	SLR	pH	Time (min)	Temp (^{0}C)	Yield (%)	Increase in yield = (b/a) ×100 = c
5HA3	30	2.5	52.5	82	3.73 (a)	439.7%
5HA6	30	1.5	52.5	82	16.4 (b)	
5HA25	30	2.5	97.5	82	2.93 (a)	938.6%
5HA7	30	1.5	97.5	82	27.5 (b)	
5HA24	50	2.5	52.5	82	3.57 (a)	784.3%
5HA18	50	1.5	52.5	82	28 (b)	
5HA9	50	2.5	97.5	54	1.23 (a)	1227.6 %
5HA13	50	1.5	97.5	54	15.1 (b)	
5HA10	50	2.5	97.5	82	4.97 (a)	495%
5HA21	50	1.5	97.5	82	24.6 (b)	

Table 2: Noticeably increased yield due to lowered pH

The other sets with an increase in yield, but not as high as those mentioned in table 2 are given in table 3. The value of c (increase in yield due to lowered pH) is considerably lower in the sets mentioned in table 3 than those in table 2, because the absolute yield at pH 1.5 is much less. This is probably due to the lower temperature (54^0C) at which the experiments are carried out. Thus, a temperature of 82^0C together with a pH of 1.5 can result in a high pectin yield from USP.

Set	SLR	pH	Time (min)	Temp (^{0}C)	Yield (%)	Increase in yield = (b/a) ×100 = c
5HA16	30	1.5	97.5	54	3.78 (b)	182.6%
5HA20	30	2.5	97.5	54	2.07 (a)	
5HA15	50	1.5	52.5	54	2.68 (b)	157.7%
5HA14	50	2.5	52.5	54	1.7 (a)	

Table 3. Decrease in yield due to change in pH from 1.5 to 2.5

d. The effect of SLR on the pectin yield was found to vary with pH. At a pH of 1.5, increase in SLR from 30 to 50 (ml of extractant solution per gram of powered USP) was found to be increasing the yield slightly. The increase in yield when the SLR is increased to 50 from 30 is not as high as in the case of the other conditions such as pH, duration of heating, temperature etc. because those conditions are more limiting than SLR (within the range studied). The increase in yield of 5HA18 may be because of the higher temperature (82^0C) at which the experiment was carried out. Also, the reason for the higher yield (15.1%) of 5HA13 (at SLR = 50) may be the increased time period of heating of the extraction medium.

At a pH of 2 or above, it was found that the yield decreases slightly as the SLR increases. This must be because of the dilution of protons in the extractant medium. At a higher pH the concentration of protons in the extractant decreases. This, when coupled with increased volume of the extractant solution, results in still lesser concentration of protons affecting the removal of pectin from other cell wall components. But this effect of dilution of protons at higher pH and SLR is solved to a certain extent when both the time of heating and temperature are. This is also the reason for the result of comparison between 5HA3 and 5HA24. These observations clearly indicate that the effect of dilution of the solution can be overcome by heating the extractant medium for a prolonged time at a higher temperature.The solid : liquid ratio should be as high as possible because as the volume of the liquid increases, more of it should be evaporated or more alcohol should be used to precipitate the pectin present in it. Also, it consumes more acid to prepare the liquid of desired pH. Therefore, it is essential to know the minimum possible volume of the liquid extractant which can extract the maximum amount of pectin from the raw material.

Estimation as calcium pectate

The percentage of calcium pectate obtained is 56.871 %. During the formation of calcium pectate, adjacent polygalacturonic chains are cross-linked by Ca^{2+} ions[21]. In this work, the percentage of calcium pectate obtained is far less than the value given for purified galacturonic acid (about 110%) [20], probably because many of the galacturonic acid residues in pectin may be methyl esterified and not available for cross-linking. Also, the crude pectin extract may contain not only galacturonic acid but other components such as galactose, arabinose, rhamnose, etc. all of which are not taking part in the formation of calcium pectate.

Degree of esterification

The DE was found to be 62.5 % by the titrimetric method. Therefore, pectin obtained from stem pith of banana is high methyl pectin.

IR analysis

The FT IR spectrum obtained is given in figure 1.

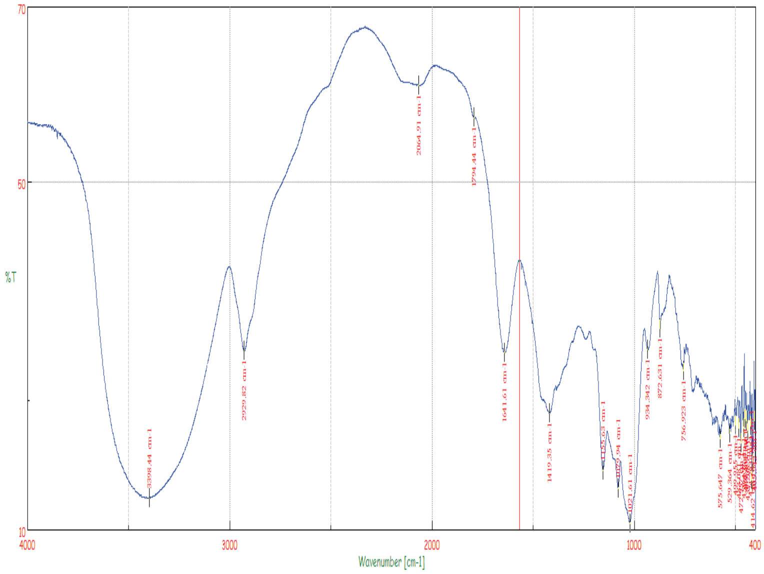


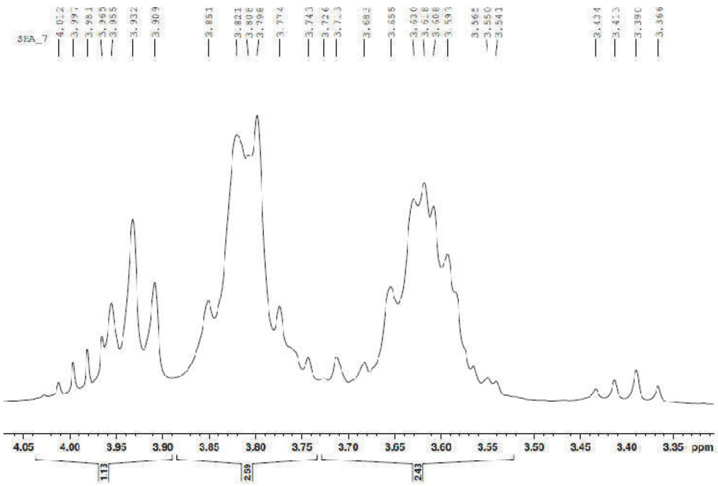
Figure FT IR spectrum of pectin extracted from stem pith of banana

The peak at 3398 cm^{-1} indicates that it contains –OH groups, commonly present in carbohydrates[22]. The peak at 2929 indicates C-H stretch, while 1794 indicates a C=O stretch. Peak at 1641 indicates N-H bend of an amide group and 1419 shows a C-H bend. The peak at 1079 represents the stretching of the bond between C and O in a methoxyl group (CH_3-O-)[22]. It was already reported that the region with strong absorption between 1200 and 950 cm^{-1}, called fingerprint region is characteristic for each type of polysaccharide and even though difficult to interpret, is independent of the source of pectin and may be instrumental in the identification of galacturonic acid[23]. Presence of these functional groups indicates that the crude extract contains pectin-like substances.

NMR analysis

The ^{1}H NMR of the pectin extracted from USP is given in fig 2. The groups/linkages identified from the spectrum are given in table 4[24]. It is seen from the ^{1}H spectrum that β- D galactose molecules are linked by means of β 1→ 4 glycosidic linkages while β 1→6 linkages are also seen. Linkages between α-D-Galacturonic acid and α-L-Rhamnose are visible, demonstrating that rhamnogalacturonans, as reported in the case of pectin from other sources,[25],[26] are present in banana pectin also. Methyl esterified galacturonic acid is visible in the spectrum.

Figure 1H NMR of pectin extracted from stem pith of banana



The ^{13}C NMR of the pectin obtained is given in fig 3. The groups identified from the spectrum are given in table 10[24].

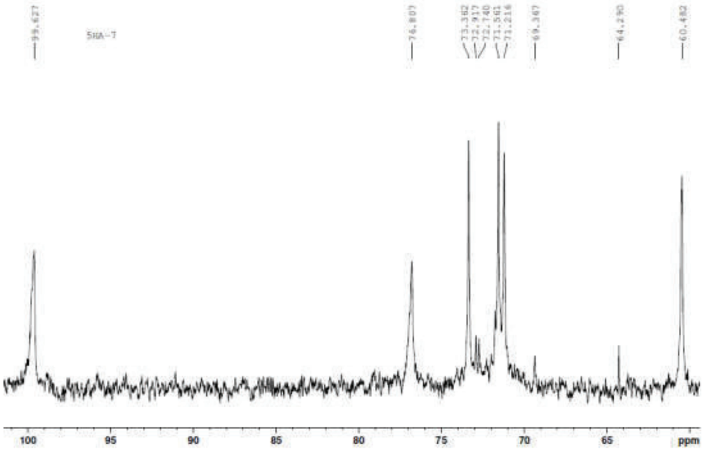


Figure ^{13}C NMR of pectin extracted from stem pith of banana

α-D-Galacturonic acid is found to be in a 1→2 glycosidic bond with α-L-Rhamno pyranose, while rhamnose – rhamnose bonds are also seen.

Sl. No	Groups identified	^{1}H Chemical shift
1	O-Me on GalpA	3.36
2	H-2 of Galp	3.39
	H-4 of →2) - α-L- Rhap-(1→(Rha)	3.413
3	H-4 of → 2) -α - Rhap-(1 →	3.43
4	H-2 of GalpA	3.63
5	H-3 of β-Galp-(1→	3.65
6	H-5 of β-D Galp - (1→4)-β-D Galp	3.68
7	H-5" of α-L-Araf-(1→(Ara)	3.71
8	H-5 of → 4)- β-DGalpOH	3.72
9	H-3 of →3)-β-D-Galp-(1→(G)	3.74
10	H-2 of →4)α-D-GalpA-(1→2) - α-L-Rhap-(1→ (GA)	3.77

11	H-5 of α-L-Araf-(1→(Ara)	3.82
12	H-3 of →2) - α-L- Rhap-(1→(Rha)	3.85
13	H-6" of → 4)- β-DGalpOH	3.9
14	H4 of β-Galp-(1→	3.93
15	H-3 of α-L-Araf-(1→(Ara)	3.96
16	H-2 of β-D Galp - (1→6)-β-D Galp	3.54
17	H-2 of β-D Galp - (1→4)-β-D Galp	3.56
18	H-2 of → 4)- β-DGalpOH	3.59
19	H-3 of β-D Galp - (1→6)-β-D Galp	3.65
20	H6 of β-Galp-(1→	3.72
21	H-3 of → 4)- β-DGalpOH	3.77
22	H-6 of β-D Galp - (1→6)-β-D Galp	3.79
23	H-5" of →5)-α-L-Araf-(1→(Ara)	3.8
24	H-6" of β-D Galp - (1→6)-β-D Galp	3.9
25	H-4 of β-D Galp - (1→6)-β-D Galp	3.95
26	H-3 of →4)α-D-GalpA-(1→GA	3.98
27	H3 of → 4)-α-GalpA(Ome)-(1→	4.01
28	H-1 of GalpA	4.71
29	H-1 of →4)α-D-GalpA-(1→2) - α-L-Rhap-(1→ (GA)	5.37

Table 4: Chemical Shifts of Proton NMR of pectin sample - 5HA7

Based on the assignment of the peaks, the following types of fractions may be identified in the extracted pectin:a. a region rich in α-D-galacturonic acid and α-L-rhamnose; some of the galacturonic acid residues are methylated [→4-α-D-GalpA(O-Me) -1→2-α-L-Rhap-1→Rha-]b. a region predominated by β-D-galactose in the pyranose form; some of the residues are substituted at C-3 [→4-β-

D-Galp-1→4- β-D-Galp-1→6- β-D-Galp→4- β-D-GalpOH] c. a region of α-L-arabinose residues

Sl. No.	Group identified	^{13}C Chemical shift
1	C-2 of →4)α-D-GalpA-(1→2) - α-L-Rhap	**69.3**
2	C-5 of →2) - α-L- Rhap-(1→(Rha)	**71.2**
3	C-3 of →4)α-D-GalpA-(1→2) - α-L-Rhap	**71.5**
4	C-2 of →3)-β-D-Galp-(1→(G)	**72.7**
5	C2 of →4)-β-D-Galp-(1→4 (G)	**73.3**
6	C-5 of →3)-β-D-Galp-(1→(G)	**76.8**
7	C-5 of →4)α-D-GalpA-(1→GA	**72.9**
8	C-5 of β-D-Galp - (1→4)-β-D Galp	**76.8**
9	Non-esterified α-D-GalA	**99.6**

Table 5 Chemical Shifts of 13C NMR of pectin sample - 5HA7

Conclusion

India, being the largest producer of banana, has tremendous potential to produce a large quantity of pectin. As shown by this study, the stem pith contains up to 28% (by dry weight) of HM (with more than 50% of the galacturonic acid residues esterified with methanol) pectin. Pectin yield increases as the temperature and duration of heating increase at a pH = 1.5, but a higher volume of the extractant solution decreases the yield at a pH of 2 or more. However, this effect may be overcome by increasing the temperature and time of heating. This study clearly indicates that stem pith of banana is a good source of HM pectin which opens up the possibility of large-scale production of pectin from banana as our country is the largest producer of banana in the world. If small scale industries are established in the rural agriculture-based communities of the country, banana plants damaged due to climatic conditions and after harvesting can be used for the production of pectin. This can make agriculture profitable, increasing the financial stability of the farmer, thus modern educated youth can be attracted in to the field of agriculture. This can also lead to the establishment of more agro-based industries nearby the farmland itself because of the availability of the raw material. Thus, more employment opportunities can be generated. In a country like India, where the majority of people depend upon agriculture for their livelihood, this kind of developments will become a necessity.

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